

SPECTRAL CHARACTERISTICS OF THE LIGHT BAND ON RED CLOVER (*TRIFOLIUM PRATENSE* L.)

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FELDHAKE C. M. *Spectral characteristics of the light band on red clover* (*Trifolium pratense* L.). ENVIRONMENTAL AND EXPERIMENTAL BOTANY **30**, 111–115, 1990. --One striking characteristic of red clover (*Trifolium pratense* L.) is the reflective band across the mid-region of the leaves of many plants. The objective of this study was to determine the nature of this band as related to potential ecological significance. A radiation budget was determined for the light and dark green regions under mid-day sunlight using measurements made with a Li-Cor, LI-1800 spectroradiometer equipped with a microscopic lens. The light region reflected nearly 70% more visible radiation than the dark green region of the leaf; however, this difference amounted to only 3.4% of possible radiation reflected by a white barium sulfate standard. There was little difference between the two regions in reflectance from the ultraviolet or infrared. The light region essentially disappeared when subjected to vacuum hydration; this result supported the hypothesis that the scattering of light was due to cell wall–air cavity interfaces. The reflective band, even when broad, does not appear to have a substantial impact on the energy budget of red clover leaves. The nature of the band and its variability within populations lend support to the argument that it is cryptic.

Key words: Solar reflectance, energy budget, cryptic.

INTRODUCTION

THE spectral characteristics of leaves are important parameters which influence how a plant interfaces with its environment. Light absorption provides the mechanism for energy capture needed in photosynthesis and is a major source of energy for transpiration which facilitates nutrient uptake.

GATES *et al.*⁽¹⁰⁾ discussed the higher reflectivity of desert plants compared to plants from more mesic regions. Desert plants are normally thicker, which reduces light transmission. In regions with generally high solar radiation and low water availability, higher reflectivity helps ameliorate the increased radiation absorption characteristic of thicker leaves.

Under moderately droughty conditions, yields of barley, sorghum and wheat^(1,6,13,16) were increased for genetic iso-lines with a white epicuticular wax. The yield increases were attributed to a higher water use efficiency and to maintenance of a higher leaf area index under water stress. MULROY⁽¹⁴⁾ hypothesized for *Dudleya brittonii* Johansen that the high reflectance of glaucous leaves is ecologically significant by reducing damage to dehydrated leaves from visible and u.v.-B radiation, thus promoting longevity of the leaves.

Leaf pubescence also increased radiation reflectance, and thereby reduced water use and leaf temperature, for the desert shrub *Encelia farinosa* Gray.^(8,9,19) Pubescence in soybeans resulted in only minor increases in reflectivity,^(12,15) with

mixed effects on water use efficiency.⁽⁷⁾ Pubescence in soybean appears to be primarily related to pest resistance.⁽¹⁷⁾

The reflectance of cotton leaves increased in response to an increase in soil salinity and to increases in leaf water deficit.^(11,21) The mechanism of increased reflectance appeared to be an increase in intercellular air space and the subsequent increase in scattering of light by cell wall-air cavity interfaces.

Red clover (*Trifolium pratense* L.) is a desirable pasture legume in much of the U.S. One remarkable characteristic of this species is the presence of a highly reflective band across the mid-leaf section of some plants. The size of this band is highly variable in most populations, ranging from non-existent to more than half of the leaf surface area. The heritability has been determined for the similar light band in white clover (*Trifolium repens* L.)^(2,4) and for subterranean clover (*Trifolium subterraneum* L.).⁽²⁰⁾

CAHN and HARPER⁽³⁾ speculated that the purpose of the light band may be to disrupt the visual outline of the leaf as protection from grazers. Their work showed that white clover populations which were heavily grazed had an increase in the per cent of the population with light bands. Contrary results were found, however, by CHARLES.⁽⁵⁾ The physical nature of this light band, however, has not been examined.

The purpose of this study was to analyze the spectral properties of the light band frequently found on the leaves of red clover plants with the hope of gaining insight into its ecological purpose. Understanding the nature of this reflective region may lead to the development of varieties of red clover better suited to specific environments.

METHODS AND MATERIALS

Several red clover plants, which had wide, pronounced, light bands, were collected from an old pasture in southern West Virginia and from a population of the variety Kenstar. The plants were grown in a greenhouse for several months to give large plants with blemish-free leaves. Also grown in the greenhouse were two grasses for comparison purposes, *Panicum amarulum* Hitchc and Chase, which was very glaucous and *Panicum virgatum* L. which was lacking glaucous wax.

Spectral measurements were made in the 300–1100 nm range with a Li-Cor LI-1800 portable spectroradiometer. The spectroradiometer was equipped with a microscopic lens. The lens configuration used was capable of measuring light from a 4-mm diameter region. Measurements were made on cloud-free days within 2 hr of solar noon. Reflected radiation measurements were made of leaves mounted on a flat plate suspended in front of the lens at the focal distance (about 10 cm) with a rigid bracket. The plate was covered with black cloth to minimize transmittance from behind. Reflection from leaves was measured about 30° from normal with incident solar radiation normal to the surface.

Transmission measurements were made by fastening leaves to a wire hoop and placing the hoop between the sun and the lens. The top leaf surface was placed normal to the sun and the reading was taken about 30° from normal through the leaf.

Close microscopic inspection suggested that the reflective mechanism was a result of an abundance of air-cell membrane interfaces near the leaf surface. This hypothesis was supported by vacuum hydration which caused the reflective band region to become indistinguishable from the darker green regions. Leaves were placed in a beaker of water and submitted to a vacuum sufficient to cause boiling at room temperature for about 5 min. Within a couple of minutes after placing hydrated leaves in the sun, the light band would rapidly return with visually no apparent short-term damage to the leaf (the leaf would obviously suffer long-term damage from being picked).

While reflection measurements were possible for hydrated leaves mounted on the plate surface it was not possible to obtain good transmission data from the vacuum hydrated leaves. With the leaves mounted on the wire hoop both sides were exposed to the wind and the reflective band would begin returning before a complete scan could be made.

RESULTS AND DISCUSSION

The solar radiation reflected from a barium sulfate coated disc, used as a white standard, is

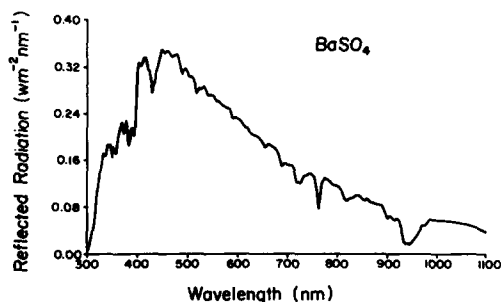


Fig. 1. Reflected radiation in the 300–1100 nm region for a white barium sulfate disc.

shown in Fig. 1. This represents the maximum reflected radiation possible under the measurement procedures used in this study. The incident solar radiation levels may have varied by a few per cent between figures but all scans on single figures were made within several minutes of each other and therefore incident radiations would not have changed substantially.

The shade of green, the brightness of the light band, and the width of the light band were all highly variable within the populations from which plants were selected. While measurements of leaves from many plants were made, only scans from leaves of a single clover plant will be shown to facilitate comparisons between figures. The plant whose characteristics are shown had a distinct broad light band and dark green color. Figure 2 shows the spectral reflected radiation of the light region and the green region of a single clover leaf. It is evident from Fig. 2 that the light band's increased reflectivity is primarily in the visible

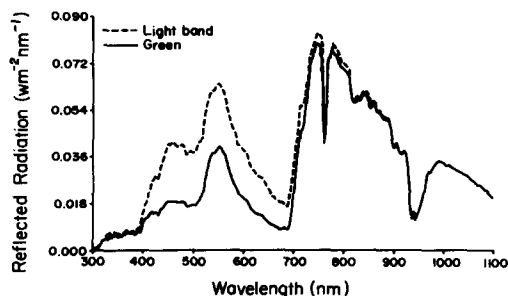


Fig. 2. Reflected radiation in the 300–1100 nm range for the light band and dark green region of a red clover leaf.

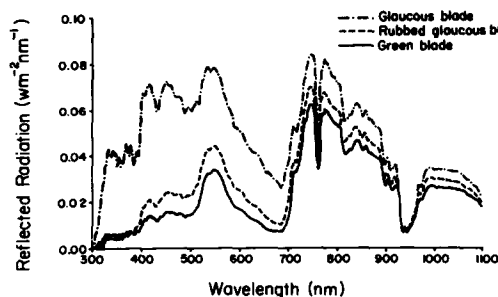


Fig. 3. Reflected radiation in the 300–1100 nm for a highly glaucous grass blade, a grass blade with most of the glaucous coating rubbed off, and a glaucous-free grass blade.

region. The band does not effectively reflect in the ultraviolet range and very little in the infrared region.

The reflected solar radiation from a glaucous and non-glaucous grass leaf is shown in Fig. 3 along with reflected radiation from a glaucous leaf with most of the white wax rubbed off using a cotton ball. The glaucous trait resulted in a high degree of reflectance in the ultraviolet region with the relative increase in reflectance due to glaucousness gradually decreasing with increasing wavelength. The glaucous trait has potential for having a much greater impact on the overall energy budget of the grass leaf than the light band does on the clover leaf, not only because of its superiority as a reflective mechanism but because it covers the entire leaf.

The transmitted radiation through both the light band and the green region is shown in Fig. 4. The transmitted radiation pattern for the green

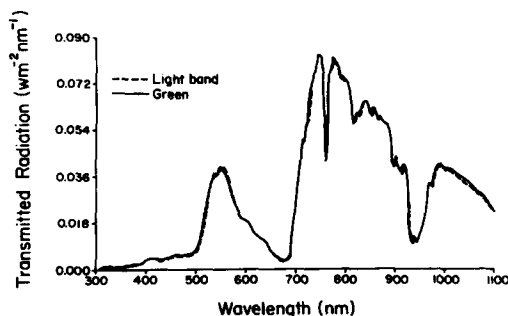


Fig. 4. Transmitted radiation in the 300–1100 nm for the light band and dark green region of a red clover leaf.

Table 1. Reflectance of clover and grass leaves as a per cent of reflectance of a white barium sulfate disc measured under the same conditions

	Per cent reflected		
	300-400 nm	400-700 nm	700-1100 nm
<i>Clover</i>			
Light band	1.8	8.3	41.9
Green	1.5	4.9	40.9
Difference (Δ)	0.3	3.4	1.0
<i>Grass</i>			
Glaucous	21.7	21.9	57.4
Green	3.4	6.3	41.4
Difference (Δ)	18.3	15.6	16.0

leaf area is very similar to the reflected pattern except for the 400–500 nm range where it is much lower. What is most remarkable, however, is that the transmitted values for the light band and the green region differ very little. This is readily evident visually. On some plants it is not possible to discern the border of the light band when viewing through the leaf from underneath in sunlight while on others it is noticeable but faint.

The radiation energy budget for a leaf can be written

$$R_i = R_r + R_t + R_a \quad (1)$$

where R_i is incoming radiation, R_r is the reflected radiation, R_t is the transmitted radiation, and R_a is the absorbed radiation. It is evident from Figs 2 and 4 that since the light band has very little influence on the amount of transmitted radiation, and assuming R_i is constant, the increase in reflection must be offset by an equal decrease in absorption.

Table 1 lists the per cent of radiation reflected in the ultraviolet, visible, and infrared for the clover and grasses. It was calculated as a per cent of the amount reflected from the barium sulfate. The glaucous grass had over 15% greater reflectance in all three regions than the green grass. The light band on the clover leaf, however, had little effect on reflectance in the ultraviolet or infrared. In the visible region the light band reflected nearly 70% more radiation than the green leaf area; however, this was only 3.4% of the maximum possible reflected radiation. Since

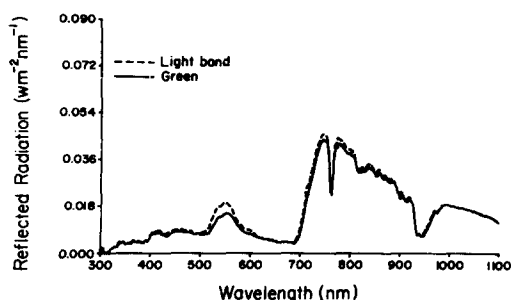


Fig. 5. Reflected radiation in the 300–1100 nm region for the light band and dark green region of a red clover leaf after being subjected to vacuum hydration.

the increase in reflection is nearly equal to the decrease in absorption, the light area absorbs only about 3.4% less visible radiation than the green area.

The light band essentially disappears when a leaf is subjected to vacuum hydration. The reflected radiation from the light band and the green area of hydrated leaves are shown in Fig. 5. The amount reflected at all wavelengths decreased as a result of this hydration process. While it was not possible to measure the radiation transmitted by hydrated leaves, visual observation indicated that the decrease in reflection was balanced by an increase in transmission which is the same effect seen when viewing a wet piece of paper.

While the light bands on the leaves of some clover plants are visually prominent, the increased reflection of that region does not appear to have a substantial effect on the energy budget of the light region itself, much less on the leaf as a whole. The increased reflection is limited to the visible region unlike the glaucous coating on the grass which is an effective reflector across the spectrum.

Highly reflective surfaces have been used to repel pests such as aphids⁽¹⁸⁾ and leafhoppers.⁽²²⁾ While the hypothesis that the light band repels pests by increasing overall albedo has not been tested, it seems unlikely since the absolute amount of light reflected is so small. However, the contrast between the two regions of the leaf is visually striking. For this reason the physical nature of the light band lends support to the hypothesis of CAHN and HARPER⁽³⁾ that it breaks up an image

of leaf shape and hinders the formation of a search image by grazing animals, or that the bands make a large leaf look like a small leaf.

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